

MORPHOLOGICAL PHYLOGENETIC CONSIDERATIONS ON THE RELATIONSHIPS OF *ISODICTYA* BOWERBANK, 1864

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Isodictya Bowerbank was recently transferred from the Order Poecilosclerida to the Order Haplosclerida, based upon the hypothesis that skeletal architecture and spiculation are homologous between *Isodictya* and some genera in the haplosclerid family Niphatidae. We examined this hypothesis by determining the diagnostic morphological characters of *Isodictya*, comparing these with a selection of poecilosclerid and haplosclerid genera, which also have reticulate spongin skeletons. A phylogenetic analysis of diagnostic morphological characters such as spicule morphology, choanosomal skeletal architecture, and surface fibre ornamentation was carried out to determine the ordinal affinities of *Isodictya*. The analysis of this data set produced 21 equally parsimonious trees of 29 steps with a consistency index (C.I) of 1.000 and retention index (RI) of 1.000. Major characters that separate *Isodictya* from the haplosclerid genera include the nature of the surface skeletal outgrowths, the amount of spongin associated with these outgrowths, the absence of a paratangential skeleton, the presence of chelae, and the presence of small cigar-shaped oxeas. Results strongly suggest the retention of *Isodictya* and allied taxa *Cercidochela* and *Esperiopsis* within the Order Poecilosclerida. □ *Porifera*, *Isodictya*, *Poecilosclerida*, *Haplosclerida*, *phylogeny*.

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The order Poecilosclerida is the largest and most taxonomically difficult and unstable of all Demospongiae (Van Soest, 1984; Bergquist & Fromont, 1988; Hajdu et al., 1994a); at present there is little consensus on family composition and classification. One major debate in poecilosclerid classification has questioned the integrity of Desmacididae Schmidt, 1870 (also incorrectly known as Desmacidonidae Gray, 1872). It is now considered to be polyphyletic (Van Soest, 1984; Hajdu et al., 1994a,b) because the family contains species with monactinal and/or diactinal megascleres, with myxillid, mycalid, and microcionid skeletal architecture, and in some genera sand replaces the megascleres. *Isodictya* Bowerbank, 1864 has been traditionally placed in this family because it contains diactinal megascleres in a reticulate skeleton and possesses chelae. Recent rearrangement of the Desmacididae by Van Soest (1984), Bergquist & Fromont (1988) and Hajdu et al. (1994a), resulted in the transfer of all desmacidid genera to other poecilosclerid families, leaving *Isodictya* unassigned.

Recently Van Soest (1987) and de Weerd (1989) postulated that Desmacididae was a sister group of Haplosclerida, with the primary synapomorphy being the presence of small oxeas (100-250µm) (de Weerd, 1989). Hajdu et al. (1994a, b) subsequently transferred *Isodictya* to the order Haplosclerida arguing that the skeletal architecture is homologous to that of genera in the haplosclerid family Niphatidae. The presence of chelae in *Isodictya*, which are absent in all haplosclerids as presently defined, was considered by Hajdu et al. (1994b) to have been secondarily lost in other haplosclerids, *Isodictya* alone retaining this plesiomorphic character. A comprehensive historical overview of this problem is given in Hadju et al. (1994a) and Bergquist and Fromont (1988).

The validity of this transfer rests on the question of whether oxeas and the pattern of their reticulation in *Isodictya* and the Haplosclerida are truly homologous. Our objectives were firstly to determine the diagnostic morphological characters for *Isodictya*, and then to consider this genus in the light of other poecilosclerid genera

TABLE 1. Material examined and locality data for *Isodictya* spp., *Esperiopsis informis* Kirkpatrick, *Cercidochela lankesteri* Kirkpatrick, *Niphates* spp., *Amphimedon* spp., *Cribrorhynchia* spp.

Species	Registration Number	Locality
<i>Isodictya palmata</i>	BMNH 1830.7.3.381	NW. Atlantic
<i>Isodictya multiformis</i>	BMNH 1997.5.12.18	Ouderkraal, South Africa
<i>Isodictya</i> sp.	BMNH 1895.6.8.140	Indo-Pacific
<i>Esperiopsis informis</i>	BMNH 1997.5.12.30	Ouderkraal, South Africa
<i>Cercidochela lankesteri</i>	BMNH 1826.10.26.179	Winter Quarters, Antarctica
<i>Niphates digitalis</i>	BMNH 1928.5.12.202	Bahamas
<i>Niphates</i> sp.	0CDN 4130-X	Micronesia
<i>Amphimedon compressa</i>	BMNH 1928.5.12.921	St Thomas
<i>Amphimedon</i> sp.	0CDN 4249-C	Micronesia
<i>Cribrorhynchia vasculum</i>	BMNH 1997.3.20.1	Bahamas
<i>Cribrorhynchia</i> sp.	0CDN 4159-G	Micronesia

such as *Esperiopsis* and *Cercidochela*. Finally, a selection of reticulate spongin skeletons in haplosclerid genera were examined and compared to those in species of *Isodictya*.

MATERIALS AND METHODS

Collections were made by the authors using SCUBA unless otherwise stated. Methods of collection, preservation, histological preparation for light microscopy examination, and scanning electron microscopy, were carried out according to Bergquist & Kelly-Borges (1995). Material examined and considered in this study is listed in Table 1. Taxa for comparison with *Isodictya* and allied genera were selected on the basis of their superficial similarity to haplosclerid genera. Genera in the haplosclerid families Chalinidae, Callyspongiidae, and Oceanapiidae were not considered here as their reticulate skeletons are quite separate from the Niphatidae whose genera have been strongly compared to *Isodictya*. Abbreviations: BMNH, Natural History Museum, London; 0CDN, Specimen sample numbers for the United States National Cancer Institute shallow-water collection programme contracted to the Coral Reef Research Foundation, Micronesia. Twelve morphological characters (Table 2) were identified from direct examination of the specimens listed in Table 1. Parallelism may be quite a common feature in sponges (de Weerd, 1989), at least when simple characters such as consistency, colour and habitats are concerned. We have avoided this by excluding them from the phylogenetic analysis. Each character was scored for each species in a taxon/character data matrix (Table 3). The data

were analysed using PAUP Version 3.1.1 (Swofford, 1993). Characters were coded as unordered and multi-state and were unweighted. Wagner parsimony (Kluge & Farris, 1969) was used as it minimises evolutionary steps by making no assumptions about the direction of character changes. Analyses were performed using an exhaustive search to find the minimum length trees. *Halichondria moorei* (Halichondridae, Halichondriidae) was defined as the outgroup, data obtained from Bergquist (1970). One hundred bootstrap replicates (Felsenstein, 1985) were carried out to provide confidence estimates on groups contained in the most parsimonious trees.

RESULTS

Phylogenetic analysis produced 21 equally parsimonious trees of 29 steps with a consistency index (CI) of 1.000, a retention index (RI) of 1.000, and homoplasy index (HI) of 0.000 (Fig. 1A,B). Even under the hypothesis of polymorphisms for multiple character states, the CI was 1.000. The advantage of using multiple states with the hypothesis of polymorphism is that this procedure permits the detection of hidden homoplasies and reversions in a study at the generic level, which otherwise would be omitted by exclusively affecting isolated species in each genus (Maldonado, 1993).

Isodictya spp. form a monophyletic clade in all 21 equally parsimonious trees, with essentially the same arrangement of species in each tree (Fig. 1). *Cercidochela lankesteri* is grouped with *Esperiopsis* and *Isodictya* in 15 of the 21 reconstructions, but in the remaining 6 trees the position of *Isodictya* is unresolved with respect to *Cercidochela* and *Esperiopsis*. The overall position of *Isodictya* is stable within the poecilosclerid clade, and this is clearly separated from haplosclerid genera which also form a distinct, yet internally unresolved clade. Removing *Cercidochela* from the analysis resolves *Isodictya* spp. as a single clade that it is more closely related to *Esperiopsis informis* than to haplosclerid genera. A strict consensus tree yielded a polytomy between the different orders but *Isodictya* remains clearly separated from Niphatidae (Fig. 2).

TABLE 2. Characters and character states of *Isodictya* spp., *Esperiopsis informis* Kirkpatrick, *Cercidochela lankesteri* Kirkpatrick, *Niphates* spp., *Amphimedon* spp., *Cribrochalina* spp.

CHARACTERS 1-2. GENERAL SKELETAL STRUCTURE: 1. Body compression: a. three-dimensional b. planar; 2. General skeletal organisation: a. plumoreticulate with interstitial isodictyal reticulation, b. square-meshed reticulation; c. confused halichondroid.

CHARACTERS 3-7. FIBRE DEVELOPMENT: 3. Primary fibres: a. small square-meshed reticulation, b. large polygonal reticulation, c. fine plumose fibres, d. robust plumose fibres, e. Absent; 4. Secondary fibres: a. regular spongin-bound ladder-like fibres, b. irregular fascicular spongin-bound fibres, c. primary fibres bridged by single spicules and tracts, d. primary fibres bridged by semi-isodictyal reticulation of spicules, e. absent; 5. Mesh shape: a. small square, b. large square, c. large elongate, d. small irregular, e. absent; 6. Ornamentation associated with termination of primary fibre: a. low blunt conule, b. large spiky conule, c. plumose tuft, d. absent; 7. Spongin development in primary fibres: a. spongin joining spicules; b. spongin entirely enclosing fibres; c. absent.

CHARACTER 8. ECTOSOME: 8. Ectosome (between primary fibres): a. tangential fibres, b. ectosomal brushes, c. tangential detachable ectosome.

CHARACTER 9. MEGASCLERES: 9. Morphology: a. small hastate oxeas, uniformly thick; b. small centrally angulate and thickened fusiform oxeas; c. styles, d. large fusiform oxeas.

CHARACTER 10. MICROSCLERES: 10. Chelae: a. palmate isochelae, normal form; b. palmate isochelae, modified; c. canonochealae; d. absent.

CHARACTER 11. BIOCHEMISTRY: 11. Manzamine alkaloids: a. present; b. absent. (Magnier & Langlois, 1998)

CHARACTER 12. REPRODUCTION: 12. Viviparity: a. present; b. Absent.

Isodictya palmata (Fig. 3A-D) and *Isodictya* sp. are characterised primarily by a plumoreticulate skeleton (character 2a) (Fig. 3B) with tufted surface outgrowths (character 6a), the possession of small fusiform oxeas (character 12c) which are often angulate and thickened centrally (Fig. 3C), and palmate isochelae (character 13a) (Fig. 3D). *Isodictya multififormis* is separated from other species of *Isodictya* by the nature of the secondary fibres (character 4b->c), the differences in mesh shape and size (character

5a->b) and morphology of the palmate isochelae (character 13a->b).

Esperiopsis informis (Fig. 3L-N) and *Cercidochela lankesteri* (Fig. 3H-K) are joined in a common clade linked by the possession of an irregular anastomosing interstitial network (character 3d) (Fig. 3I,M), absent in all other taxa except *Isodictya*. These two genera form a common clade with *Isodictya* spp. sharing plumose surface outgrowths (character 6a), absence of special dermal skeleton found only in Niphatidae, and isochelae. Canonochealae (Fig. 3K) are unique to *Cercidochela lankesteri* (character 13a->c) while *Esperiopsis* has palmate isochelae (character 13a) (Fig. 3H). *Esperiopsis informis* is unique in this analysis as it has styles (character 12a->d).

TABLE 3. Character state matrix. Characters and states are described in Table 2. For certain characters some taxa may not logically possess a given state, or the authors are unsure of the character state assignment, in which case these character states are coded as 'unknown' which is indicated by '?'; * = the outgroup.

Species	Character											
	1	2	3	4	5	6	7	8	9	10	11	12
<i>Isodictya palmata</i>	b	a	c	c	c	c	a	b	b	b	b	a
<i>Isodictya multififormis</i>	b	a	c	c	d	c	c	b	b	a	b	a
<i>Isodictya</i> sp.	b	a	c	c	c	c	a	b	b	a	b	a
<i>Esperiopsis informis</i>	b	a	c	d	d	c	c	b	c	a	b	a
<i>Cercidochela lankesteri</i>	b	a	d	d	d	c	c	b	d	c	b	a
<i>Niphates digitalis</i>	a	b	b	b	b	b	b	a	a	d	a	a
<i>Niphates</i> sp.	a	b	b	b	b	b	b	a	a	d	a	a
<i>Cribrochalina</i> sp.	a	b	a	a	a	a	b	a	a	d	a	a
<i>Amphimedon</i> sp.	a	b	a	a	a	a	b	a	a	d	a	a
<i>Amphimedon compressa</i>	a	b	a	a	a	a	b	a	a	d	a	a
<i>Halichondria moorei</i> *	?	c	e	e	e	d	c	c	d	d	b	a

Niphatidae are united by several synapomorphies. They all have hastate oxeas (Fig. 3G) (character 12b->a), a para-tangential ectosome and surface conules surrounded by spongin (character 7a->b). The interstitial skeleton of Niphatidae is limited to individual spicules rather than an anastomosing network as in the *Isodictya* group. Major characters that separate *Isodictya* from Niphatidae include the nature of the surface skeletal outgrowths (plumose tufts in *Isodictya* and conules in *Niphates*), the amount of spongin associated with these outgrowths in Niphatidae, spongin being absent in

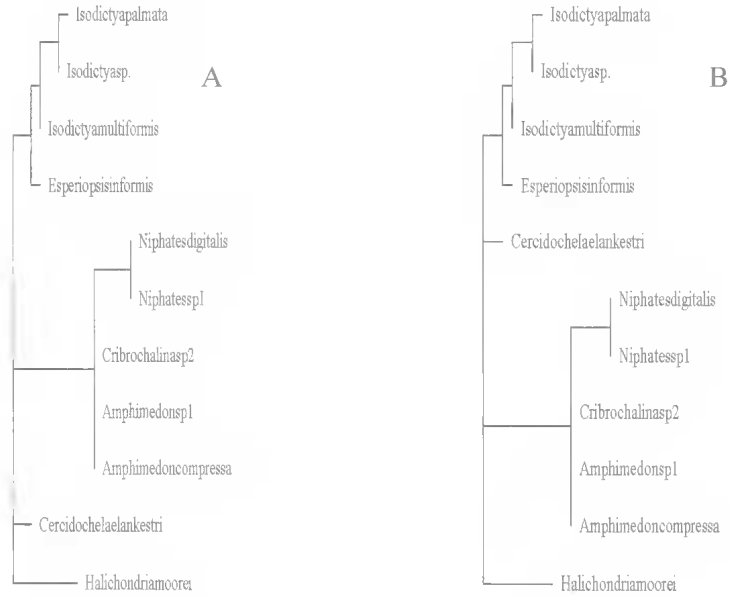


FIG. 1. A-B, Alternative, equally likely hypothetical phylogenies for *Isodictya* spp. with respect to allied genera *Cercidochela* and *Esperiopsis*, and selected haplosclerid genera.

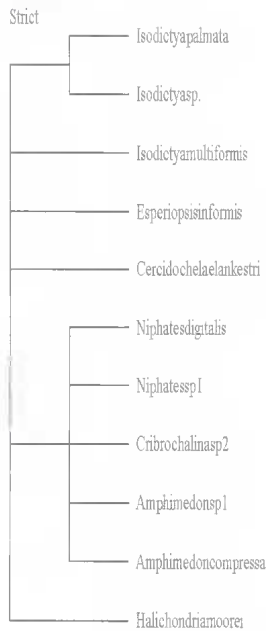


FIG. 2. Strict consensus tree of the 21 MP trees generated from the morphological data for the 12 sponge species.

Isodictya outgrowths, the presence of a paratangential ectosomal skeleton in Niphatidae, the presence of chelae in *Isodictya*, and the presence of small curved hastate oxeas of uniform thickness, in Niphatidae. A bootstrap 50% majority rule consensus cladogram (Fig. 4) provides good support for separation of the *Isodictya* group from niphatid genera, strongly suggesting that they belong to separate orders.

DISCUSSION

The diagnostic character reversal suggested by Hajdu et al. (1994b) as being synapomorphic (viz. loss of chelae) is inconsistent with the present analysis. It is more parsimonious to regard the appearance of chelae in the Poecilosclerida, rather than their loss, as a subsequent achievement in the evolution of this order. Thus, the presence of chelae is a synapomorphic character for the poecilosclerid genera under study. Moreover, although suggested as symplesiomorphic at the species level, palmate

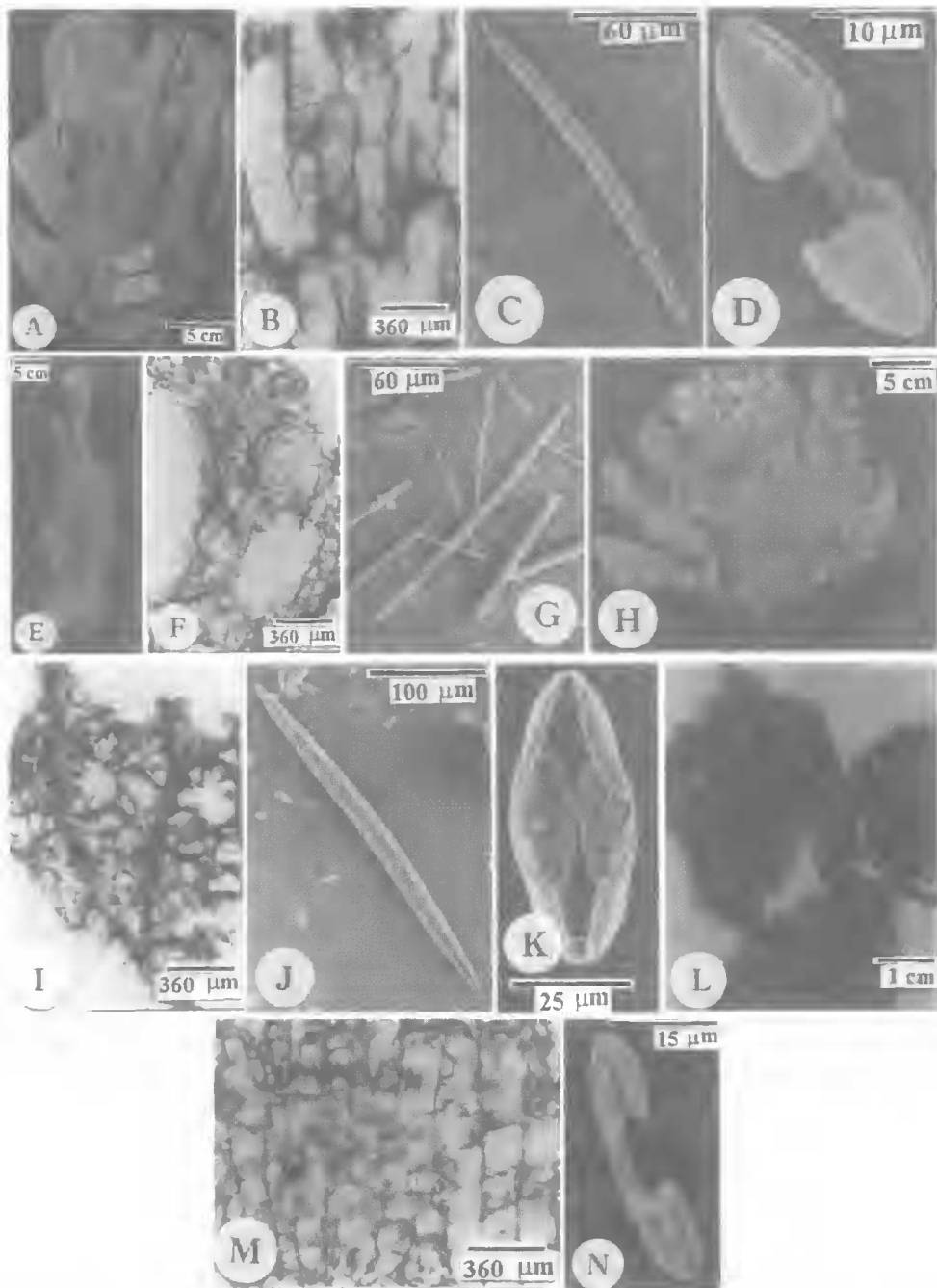


FIG. 3. Morphological characters of *Isodictya*, *Amphimedon*, *Cercidochela* and *Esperiopsis*. A-D, *Isodictya palmata* Bowerbank, 1864 (BMNH 1895.6.8.140). A, Holotype. B, Skeletal architecture ($\times 31$). C, Oxea morphology ($\times 500$). D, Profile view of palmate isochela ($\times 3,000$). E-G, *Amphimedon compressa* Duch. & Mich., 1864 (BMNH 1928.5.12.921). E, Specimen. F, Skeletal architecture ($\times 31$). G, Oxea morphology ($\times 700$). H-K, *Cercidochela lankesteri* Kirkpatrick, 1906 (BMNH 1826.10.26.179). H, Holotype. I, Skeletal architecture ($\times 31$). J, Oxea morphology of ($\times 300$). K, Profile view of canonochela ($\times 3,000$). L-N, *Esperiopsis informis* Stephens, 1915 (BMNH 1997.5.12.30). L, Specimen. M, Skeletal architecture ($\times 31$). N, Profile view of palmate isochela ($\times 3,000$)

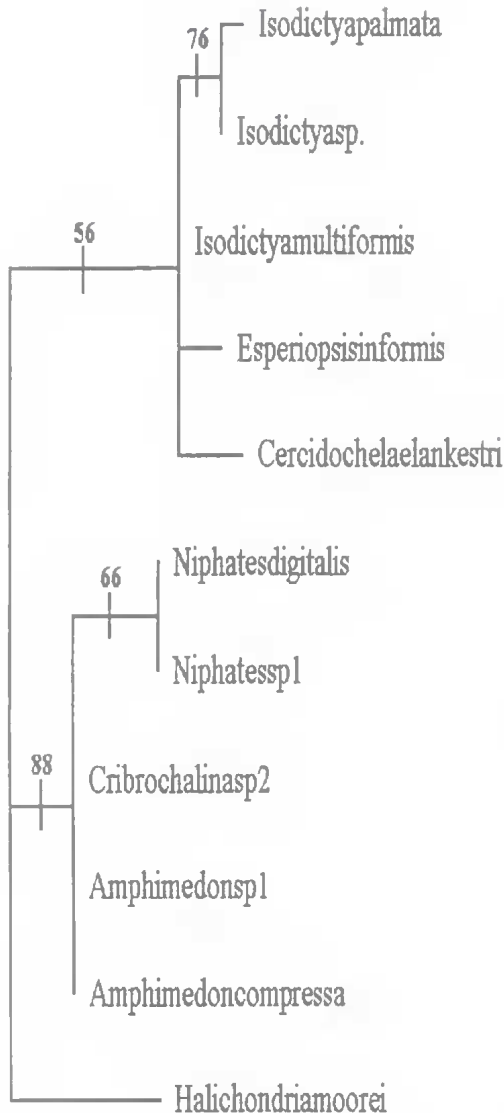


FIG. 4. Bootstrap 50% majority rule tree of the 21 MP trees generated from the morphological data for the 12 sponge species. The percentage of these trees that contain each component is shown along each branch.

isochelae can be treated as a synapomorphy when discussing generic or familial affinities (Hajdu et al., 1994b).

The similarity between the principal morphological characters of *Isodictya*, *Esperiopsis* and *Cercidochela* are striking; the independent development of the skeletal architecture, ocolourea morphology, and chelae morphology is considered to be unlikely. Hajdu et al. (1994b) based their transfer of *Isodictya* to Niphatidae

primarily on the presence of what they regarded to be an 'isodictyal' reticulation, and secondarily on the presence of palmate isochelae in *Isodictya*. These microscleres are absent in all other Haplosclerida, and are considered to be simply an underlying synapomorphy to both orders by Hadju et al. (1994a). Close examination of the reticulate skeletons of pertinent genera within these two groups shows here that niphatid skeletons are not isodictyal in the strict sense, a term that should be reserved for genera within the haplosclerid family Chalinidae, and that they are very different from the reticulate skeletons of *Isodictya*, *Esperiopsis* and *Cercidochela*. Moreover, additional characters examined here, such as the nature of surface ornamentation, megasclere morphology, interstitial spiculation, nature of the actual fibres and the exclusive presence of manzamine alkaloids (Magnier & Langlois, 1998) in the haplosclerid family Niphatidae and Chalinidae add confidence to the separation of these to groups.

Even though palmate isochelae are present in *Esperiopsis*, it is presently placed in the poecilosclerid family Mycalidae whose genera all contain anisochelae. If other skeletal characters are considered, such as the nature of the primary fibres and surface ornamentation, *Isodictya* may also fit within this family (see Hooper, 1997).

Many of these decisions on affinities may be further corroborated and illuminated with additional taxa and additional tools such as molecular systematics. Until further studies are carried out on the detailed nature of the skeletal morphology, spiculation, and chemistry of *Isodictya*, this genus should be retained within Poecilosclerida.

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